**Analyzing, Interpreting and Presenting Microbiome-Resistome Data**

Requirements

* Laptop with WiFi capabilities
* Software – pending
* Prerequisite skills, courses or experience – none
* Recommended pre-course reading and tutorials will be distributed prior to the course

At the end of the course, attendees will be able to:

* Understand features of study designs and research methods (e.g., sampling, handling, processing, etc) that promote high-quality, reproducible microbial ecology researchDescribe and interpret ecological parameters as they pertain to microbial communities.
* Understand the workflow and underlying molecular biology principles related to amplicon and shotgun sequencing, including both short- and long-read platforms.
* Master command line coding for file management and bioinformatic analysis on remote servers.
* Understand principles of different computational methods for classification of metagenomic sequencing data.
* Understand and interpret summary data describing the quality of sequencing output.
* Use a remote high-performance computing platform and local computing to conduct bioinformatic analysis of short-read metagenomic sequencing data.
  + Use Qiime and its associated software (e.g., DADA2) to generate classification tables characterizing microbial community membership/structure using 16S amplicon sequencing data.
  + Use Kraken to generate classification tables characterizing microbial community membership/structure using metagenomic shotgun sequencing data.
  + Use AMR++ pipeline and associated software to generate classification tables regarding antimicrobial resistance genes found in metagenomic microbial communities using shotgun metagenomic data.
* Identify the most widely used bioinformatic and statistical platforms being used for microbial ecology research (beyond Qiime, kraken and AMR++)
* Use R to conduct statistical analyses of metagenomic sequencing data for the following purposes:
  + Descriptive analysis of microbial features in metagenomic communities.
  + Contrasting different microbial community structures.
  + Testing hypotheses regarding microbial community composition.
  + DA testing???
* Prepare graphical figures for effective illustration of research findings from metagenomic sequencing data, and be able to interpret the figures.

Examples of research questions that people ask using the approaches you will learn in this course:

What is the microbial profile of this set of samples, and how much variability is there?

What is the resistome profile of this set of samples, and how much variability is there?

Does the microbiome or resistome profile differ between groups of samples, based on various study design factors (e.g., treatment versus control, age, diet, disease status)?

Does microbiome or resistome diversity differ between groups of samples, based on various study design factors (e.g., treatment versus control, age, diet, disease status)?

Does the relative abundance of specific types of microbes or resistance genes differ between groups of samples, based on various study design factors (e.g., treatment versus control, age, diet, disease status)?

Are there potential pathogens within the microbiome of a given (set of) sample(s)?

Are there specific AMR genes within the microbiome of a given (set of) sample(s)?

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The Microbial Ecology Group is hosting a 5-day workshop on analysis, interpretation and graphical presentation of microbiome and resistome data. Participants will learn the fundamentals of microbiome-resistome studies, from design to interpretation; and they will perform real-time, hands-on analysis of microbiome-resistome data, from raw sequence to publication-ready figures. The workshop will use a custom-built remote server pre-loaded with all necessary software packages and sequence data. This workshop has been taught dozens of times by the Microbial Ecology Group, using virtual, hybrid and in-person formats. Here are unsolicited emails we’ve received from previous participants:

“I wanted to thank you for the workshop. It was great! My main purpose was to understand the type of analysis done with metagenomics and resistome studies and I think that the workshop was a great opportunity to achieve this knowledge and beyond.”

“I would like to thank you and your team for preparing and doing so great work with the metagenomic workshop. My expectations were fulfilled in terms of getting a better general understanding of the metagenomic-tools that we have to answer scientific questions and the limitations of those tools. I believe from an epi point of view this area is a beautiful challenge and I'm willing to explore it more after this workshop.”

“It was a wonderful experience attending this workshop. I would like to use these methods in future projects back in my home country.”

“I would like to thank you for a great seminar! I thought that the lecture was well structured and that the interactive portion of the workshop was very helpful and intuitive. I left with a much better understanding of bioinformatics, the basics of metagenomics, and feel much more confident in using the programing tools to process data.”

Data analysis with long reads??

Target enrichment data analysis??

Question for Lee – can we use *only* RStudio for entire 16S analysis (not on server)?? Qiime2 or separate modules??

**Day 1 –**

**AM: Intro to microbial ecology**

**AM/PM: Conducting microbial ecology research with animal populations**

* + **Study design, sample collection and storage**

**PM: 16S – starting with library prep and sequencing**

**ASVs? OTUs? DADA2 pipeline details?**

**AM: 16S completion (up to count matrix, taxa table)**

**Day 2 –**

**PM: Shotgun/TE Starting with library prep and sequencing**

**AM: Classification of metagenomic data, including long-read**

**AM/PM: Databases, deeper dive on k-mers and alignment**

**PM: Command line work, run tutorial data**

**Day 3 –**

**Day 4 – Statistics (exploration, description and comparison) – RStudio**

**Day 5 – Interpretation and Presentation of Results – RStudio**